*Res. Rep. Agri. Sci. Tech.*
*Chungnam Natl. Univ. Korea*
*Vol. 12 (2), 163~165 (1985)*

# Effect of Acetone on the Mycelial Growth and Spore Germination of Some Phytopathogenic Fungi in vitro

**Jong Seong Park***

# 몇가지 植物病原菌의 菌絲生長과 胞子 發芽에 미치는 Acetone의 影響

朴 鍾 聲*

## SUMMARY

In assay of antifungal activity of fatty acids which slightly soluble in water against phytopathogenic fungi in vitro, it is necessary that evenly disperse fatty acid to assay in culture medium for mycelial growth or spore germination of test fungus. Acetone was selected as a dispersant in such an in vitro test because it is not only used as a solvent for fats, oils and waxes but also miscible with water. In this study to detect practically usable concentration of acetone in culture medium for test fungus, the results showed that both mycelial growth and spore germination of each test fungus were significantly inhibited on the culture media with 10% of acetone, while they were slightly inhibited on the culture media with 5% of acetone. Even though both mycelial growth and spore germination were influenced on the culture media with 5% of acetone, it is not considered that the use of culture medium with 5% of acetone make a mistake in assay of antifungal activity of fatty acids against phytopathogenic fungi.

## Introduction

The author and his co-workers reported that the extract of common purslane showed antifungal activity against some phytopathogenic fungi such as *Alternaria alternata* Japanese pear pathotype, *Pyricularia oryzae* and *Valsa ceratosperma*[1] and antifungal components in the extract were short-chain (C–C) fatty acids such as iso-butyric, n-butyric, iso-valeric, n-valeric and n-caproic acid.[2] In spite of an advantage of these fatty acids are stable to high temperature, the other fatty acids except n-butyric do not completely miscible with

* 農科大學 農學科 (Dept. of Agronomy, Coll. of Agriculture, Chungnam Natl, Univ., Taejŏn, Korea)

water.[3] It attracts authors attention that the solubility of n-Caproic acid (1.082 g/100g) in water is lowest in spite of its highest antifungal activity among those of fatty acids. It is an important task for assay of the antifungal activity that evenly disperse each fatty acid in culture medium of test fungus in vitro. Acetone well known as a solvent for fats, oils and waxes was selected as a dispersant to be used in this study.

This paper presents informations about the effect of acetone as a dispersant on mycelial growth and spore germination of some phytopathogenic fungi used for assay of antifungal activity of fatty acids in vitro.

## Materials and Methods

Culture media for test fungi: Water agar (2%) for spore germination and potato sucrose agar (PSA) for mycelial growth of test fungi were prepared respectively. No attempt was made to control pH of each medium containing a given concentration of acetone.

Concentration of acetone: Each medium containing 5% and 10% of acetone was prepared respectively as follows; Each medium was prepared respectively, autoclaved and cooled to approximately 50°C in a water bath. Then, each medium was delivered at the rate of 15 ml/plate after a given concentration of acetone was added. The control in this experiment was prepared similarly, but without the addition of acetone.

Test fungi: *Alternaria alternata* Japanese pear pathotype (Stock culture of Nagoya University), *Pyricularia oryzae* (Stock culture of Kyushu University) and *Valsa ceratosperma* (Stock culture of Chungnam University) were used as test fungi to evaluate effect of acetone against both the mycelial growth and spore germination.

Preparation of inoculum: Mycelial mats of each test fungus was prepared for the mycelial culture from the 5 to 7 day-old cultures on PSA at 26°C, except otherwise noted. Spores of each test fungus was prepared as follows; spores formed on PSA in petridishes were harvested by blushing and suspending in distilled water, each spore suspension was then sieved through a few of layers of cheese-cloth and adjusted to give the final concentration of each spore suspension which was determined microscopically to observe 20-40 spores/field at 100 X.

Culture of test fungi and observation of their mycelial growth and spore germination: After approximately 4 mm mycelial mat of each test fungus was seeded on PSA containing a given concentration of acetone, each plate was incubated for 120 to 168 hr at 26°C and mycelial growth on each plate was measured. In the other hand, after each spore suspension of test fungi was seeded on the water agar containing a given concentration of acetone by a glass atomizer, each plate was incubated for 24°hr at 26°C and rate of germination and germ-tube elongation of spores were observed microscopically.

## Results and Discussions

Effect of acetone on the mycelial growth of test fungi : All of test fungi showed significantly slow growth of mycelium on PSA containing 10% of acetone, while they showed slightly slow growth on PSA containing 5% of acetone as shown

**Table 1.** Mycelial growth of test fungi on PSA containing different concentration of acetone in vitro

| Concentration of acetone | *Alternaria alternata* Japanese pear patho-type [1] | *Pyricularia oryzae*[2] | *Valsa ceratosperma*[1] |
|---|---|---|---|
| 0 % | 37.2 mm | 26.4 | 43.2 |
| 5 % | 32.1 | 23.0 | 38.7 |
| 10 % | 24.7 | 16.3 | 29.8 |

1) Mean of 3 rep. after incubation for 5 days at 26 ± 1°C
2) Mean of 3 rep. after incubation for 7 days at 26 ± 1°C

in Table 1. Even though each test fungus was slightly inhibited in the mycelial growth on PSA containing 5% of acetone, it was considered that culture medium containing 5% of acetone may be used for assay of antifungal fatty acids which slightly soluble in water.

Effect of acetone on the rate of spore germination and germ-tube elongation of test fungi: All of test fungi showed not only significant reduction of the germination rate but also retardation of germ-tube elongation of spores on the water agar containing 10% of acetone. In the other hand, all of test fungi showed slight retardation of germ-tube elongation, while none of the test fungi showed significant differences from the control in the spore germination rate on the water agar containing 5% of acetone as shown in Table 2. It was also considered that culture medium containing 5% of acetone may be used for assay of antifungal activity of fatty acids against phytopathogenic fungi.

**Table 2.** Germination rate and germ-tube elongation of spores of test fungi on the water agar containing different concentration of acetone

| Concentration of acetone | *Alternaria alternata* Japanese pear patho-type | | *Pyricularia oryzae* | |
|---|---|---|---|---|
| | GR [1] | GTE/SL [2] | GR | GTE/SL |
| 0 % | 98 % [3] | 10.0 times [3] | 96.5 % | 10.0 |
| 5 | 94 | 8.5 | 93.0 | 7.5 |
| 10 | 59 | 6.5 | 57.0 | 6.0 |

1) Rate of germination
2) Ratio of elongated germ-tube to spore length
3) Mean of 3 rep. 5 fields/100 X.


## 摘　　要

植物病原菌에 對해서 抗菌作用을 갖고 물에 잘 녹지 않는 脂肪酸의 抗菌作用을 檢定하는데 있어서 供試菌의 菌絲生長이나 혹은 胞子發芽를 위한 培地中에 檢定하기 위한 脂肪酸을 고루 擴散시킨다는것이 必要하다. 이와같은 實驗室內의 實驗에 있어서 하나의 擴散劑로서 물과 잘 混合하며 脂肪이나 油脂나 또는 왁스 등에 溶劑로서 쓰이고 있는 아세톤을 선발했다. 供試菌에 對해 培地中에 添加하여 실제로 使用할 수 있는 아세톤의 濃度를 찾기 위한 이 硏究에서 10%의 아세톤을 含有하는 培地 위에서 各各의 供試菌의 菌絲生長이나 胞子發芽가 크게 沮害된 반면에 5%의 아세톤을 含有하는 培地에서는 약간 沮害되었다. 5%아세톤을 含有하는 培地에서 菌이 影響을 받는다 해도 5%아세톤을 含有하는 培地의 利用이 植物病原菌에 對한 脂肪酸의 抗菌作用을 檢定하는데 있어서 어떤 誤謬를 범할 것으로는 생각되지 않는다.


## REFERENCES


1. Park, J.S. et al (1984): Antifungal activity of the extract of common purslane (*Portulaca oleracea* L.). Res. Rept. of Inst. of Agr. Sci. and Tech. **11** (2): 190-193, Chungnam National University.
2. Park, J.S. et al (1985): Antifungal substances obtained from common purslane. Abstracts of the papers presented at the Annual Meeting of the Society, Fukuoka, April 2, 1985, Ann. Phytopath. Soc. Japan **51** (3): 380.
3. The Merck Index, 3rd ed., 1 (1983).